AMENDMENTS TO THE CLAIMS

1. (Original) A method for detecting genetic variation or polymorphism, i.e. a mutation, in a defensin gene comprising the steps of:

i) providing a biological sample taken from a subject to be tested,

ii) detecting the presence or absence of a variant genotype of the defensin gene in the biological sample, the presence of a variant defensin genotype indicating an increased risk of cardiovascular disease in said subject.

2. (Original) The method according to claim 1, wherein said variant genotype of the defensin gene is a homo- or heterozygote form of the mutation.

3. (Original) The method according to claim 1, wherein the detection step is a DNA-assay.

4. (Original) The method according to claim 1, wherein the detection step is carried out using a gene or DNA chip, microarray, strip, panel or similar combination of more than one genes, mutations or RNA expressions to be assayed.

5. (Original) The method according to claim 1, wherein the allelic pattern is determined using polymerase chain reaction.

6. (Original) The method according to claim 1, wherein the biological sample is a blood sample or buccal swab sample and genomic DNA is isolated from said sample.

7. (Original) The method according to claim 1, wherein the detection step is based on a capturing probe.

8. (Original) The method according to claim 1, wherein said method is used for determining whether a subject will benefit from treatment with a drug, nutrient or other therapy enhancing the defensin production, levels or activity or inhibiting defensin catabolism or elimination in the subject.

9. (Original) The method according to claim 1, wherein said method is used for determining whether a subject will be at increased risk of adverse effects or reactions if defensin antagonists are administered to a subject.

10. (Original) The method according to claim 1, further comprising a step of selecting a subject with a defensin gene sequence reducing the expression, production or levels of defensin protein for clinical drug trials testing the anticoronary and myocardial ischaemia preventing effects of compounds.

11. (Original) The method according to claim 1, wherein said cardiovascular disease is acute myocardial infarction (AMI) or coronary heart disease (CHD).

12. (Currently Amended) The method according to ~~any one of the previous claims~~claim 1, wherein said defensin is selected from the group consisting of: beta-defensin-1, beta-defensin-129, and alfa-defensin-5.

13. (Original) The method according to claim 12, wherein said variant genotype is human beta-defensin-1 gene comprising 3`UTR +5A→G mutation.

14. (Original) The method according to claim 12, wherein said variant genotype is human alfa-defensin-5 gene comprising IVS1 +198C→T mutation and/or IVS1 +243G→C mutation.

15. (Original) The method according to claim 12, wherein said variant genotype is human beta-defensin-129 gene comprising IVS1-13_12insCTC mutation.

16. (Original) The method according to claim 1, wherein genetic variation is further determined from the genes selected from the group consisting of:

a) alpha$_{2B}$-adrenoceptor,

b) apolipoprotein B, and

c) beta-2-adrenergic receptor

wherein the presence of a variant genotype in said genes indicates an increased risk of cardiovascular disease in said subject.

17. (Original) The method according to claim 16, wherein said variant genotype is alpha-$_{2B}$-adrenoceptor gene comprising insertion/deletion mutation, or said variant genotype is beta-2-adrenergic receptor comprising Gly16Arg and/or Glu27Gln mutation.

18. (Original) The method according to claim 16, wherein said variant genotype is apolipoprotein B gene comprising Thr98Ile mutation.

19. (Currently Amended) The method according to ~~any one of the previous claims~~claim 1, further comprising a step of combining information concerning age, gender, the family history of hypertension, diabetes and hypercholesterolemia, and the medical history concerning cardiovascular diseases or diabetes of the subject with the results obtained from step ii) of the method for confirming the indication obtained from the detection step.

20. (Original) The method according to claim 19, wherein said information is about hypercholesterolemia in the family, smoking status, use of cholesterol lowering medications, CHD in the family, history of cardiovascular disease, obesity in the family, and waist-to-hip circumference ratio (cm/cm)

21. (Currently Amended) The method according to claim ~~19~~ 1, further comprising a step determining blood, serum or plasma cholesterol, HDL cholesterol, LDL cholesterol, triglyceride, apolipoprotein B and AI, fibrinogen, ferritin, transferrin receptor, C-reactive protein, serum or plasma insulin concentration.

22. (Currently Amended) The method according to claim ~~19~~ 12, wherein the detected mutations are 3'UTR+5 A/G of the beta-defensin-1 gene, an insertion/deletion of three glutamic acids in the region of 12 Glu aminoacids in the codons 298-309 of Alpha-$_{2B}$-adrenoceptor, and the Thr98Ile of apolipoprotein B gene.

23. (Currently Amended) The method according to claim ~~19~~1 further comprising a step of calculating the probability of a cardiovascular disease using a logistic regression equation as follows:

$$\text{probability of a cardiovascular disease} = [1 + e^{(-(-a + \Sigma(bi*Xi))}]^{-1}$$

where e is Napier's constant, X_i are variables related to the cardiovascular disease, b_i are coefficients of these variables in the logistic function, and a is the constant term in the logistic function.

24. (Original) The method according to claim 23, wherein a and b_i are determined in the population in which the method is to be used.

25. (Original) The method according to claim 23, wherein Xi are selected among the variables that have been measured in the population in which the method is to be used.

26. (Original) The method according to claim 23, wherein b_i are between the values of –20 and 20 and/or wherein X_i are binary variables that can have values or are coded as 0 (zero) or 1 (one).

27. (Original) The method according to claim 23, wherein i are between the values 0 (none) and 100,000.

28. (Original) The method according to claim 23, wherein subject's short term, median term, and/or long term risk of CHD and/or AMI is predicted.

29. (Original) A method for targeting the treatment of CHD and AMI in a subject with CHD by determining the pattern of alleles encoding a variant defensin, i.e. by determining if said subject's genotype of the defensin is of the variant type, comprising the steps presented in claim 1, and treating a subject of the variant genotype with a drug affecting defensin production or metabolism of the subject.

30. (Original) The method according to claim 29, wherein said defensin is as defined in claim 12.

31. (Original) The method according to claim 30, wherein the variant genotype is as defined in any one of claims 13-15.

32. (Currently Amended) The method according to claim any one of ~~calims 29-31~~claim 29, wherein said variant genotype of the defensin is a homozygote or heterozygote form of mutation.

33. (Currently Amended) The method according to ~~claims~~ claim 29, wherein said CHD is angina pectoris or other form of CHD.

34. (Currently Amended) A method for treating a human or animal suffering from CHD or AMI, said method comprising:

providing said human or animal with a therapy, wherein said therapy ~~enhancing~~enhances defensin availability, enhances defensin production or enhances defensin concentration ~~of~~in the human subject or animal.

35. (Original) The method of claim 34, wherein said animal is a mammal.

36. (Original) A method for treating vascular complications of CHD and AMI, said method comprising a step of enhancing defensin availability, production or concentration in the circulation of a human subject or animal.

37. (Original) The method according to claim 34 or 36, wherein said defensin is as defined in claim 12.

38. (Currently Amended) The method according to claim ~~37~~34 or 36, ~~said method comprising administering to a subject a compound enhancing~~ wherein said defensin is Beta-defensin-1 ~~availability, production or concentration of the subject~~.

39. (Currently Amended) The method according to claim 34 or 36, wherein the said method of treating is selected from at least one of the group consisting of: a dietary treatment ~~or~~, a vaccination, gene therapy and gene transfer.

40. (Cancelled)

41. (Currently Amended) The method according to claim ~~40~~39, wherein said therapy comprises the transfer of the non-variant Beta-defensin 1 gene or fragment or derivative thereof.

42. (Original) A kit for detecting genetic variation or polymorphism, i.e. a mutation, in a defensin gene for the determination of a risk of acute myocardial infarction, AMI, and coronary heart disease, CHD, in a subject, comprising means for defensin gene allele detection, and optionally software to interpret the results of the determination.

43. (Original) The kit according to claim 42, wherein said defensin is as defined in claim 12.

44. (Original) The kit according to claim 42, wherein genetic variation or polymorphism, i.e. a mutation, is further detected in the genes selected from the group consisting of:

a) alpha$_{2B}$-adrenoceptor

b) apolipoprotein B, and

c) beta-2-adrenergic receptor.

45. (Original) The method according to claim 44, wherein the genetic variation to be detected is as defined in any one of claims 13-15.

46. (Currently Amended) The kit according to claim ~~45~~42 comprising a capturing nucleic acid probe specifically binding to the variant genotype as defined in any one of claims 13-15.

47. (Currently Amended) The kit according to ~~any one of claims 42-46~~ claim 42, comprising a DNA chip, microarray, DNA strip, DNA panel or real-time PCR based tests.

48. The kit according to ~~any one of claims 42-47~~ claim 42, comprising a questionnaire for obtaining patient information concerning age, gender, height, weight, the family history of hypertension and hypercholesterolemia, the medical history concerning cardiovascular diseases.

49. (Original) An isolated variant nucleic acid encoding alfa-defensin-5 protein, said nucleic acid comprising IVS1 +198C→T and/or IVS1 +243G→C mutation.

50. (Original) An isolated variant nucleic acid encoding beta-defensin-129 protein, said nucleic acid comprising IVS1 –13_12 in/del CTC mutation.

51. (Original) The nucleic acid according to claim 49 or 50, wherein said nucleic acid is a genomic nucleotide sequence.

52. (Original) The nucleic acid according to claim 51, wherein said nucleic acid is cDNA.

53. (Original) The nucleic acid according to claim 49 or 50 comprising an RNA sequence.

54. (Original) The nucleic acid according to claim 49 having the nucleic acid sequence set forth in SEQ ID NO:7.

55. (Original) The nucleic acid according to claim 50 having the nucleic acid sequence set forth in SEQ ID NO:32.

56. (Original) A capturing probe binding to the nucleic acid according to claim 49 or 50.

57. (Currently Amended) ~~The~~ A capturing probe ~~according to claim 56~~binding to the nucleic acid according to claim 49 or 50, ~~which comprises a single strand of the cDNA according to claim 52~~wherein said probe comprises a single strand of genomic nucleotide sequence and said genomic nucleotide sequence is cDNA.

58. (Currently Amended) The capturing probe according to claim 56 ~~or 57~~, which ~~is~~ specifically ~~binding~~binds to a variant defensin nucleic acid ~~according to claim 49 or 50~~ encoding

alfa-defensin-5 protein, said nucleic acid comprising IVS1 +198C→T and/or IVS1 +243G→C mutation, or

beta-defensin-129 protein, said nucleic acid comprising IVS1 –13_12 in/del CTC mutation,

~~but~~ wherein said capturing probe ~~do~~does not bind non-variant defensin.

59. (Original) A method for determining the presence or absence of a nucleic acid as defined in claim 49 or 50 in a biological sample comprising the steps of:

a) treating said sample to obtain single stranded target nucleic acid, or if the target nucleic acid are already single stranded, directly employing step (b);

b) contacting said target nucleic acid with a capturing nucleic acid probe and a detector nucleic acid probe;

c) detecting the complex of capturing probe, target nucleic acid and detector probe.

60. (Currently Amended) The method according to claim 59, wherein the capturing nucleic acid probe is attached or capable of attaching to a solid phase, and ~~comprises the cDNA sequence according to claim 52~~wherein said probe comprises a single strand of genomic nucleotide sequence and said genomic nucleotide sequence is cDNA, and wherein a detected signal from the solid phase is an indication of the presence in the sample of a nucleic acid ~~as defined in claim 49 or 50~~ encoding

alfa-defensin-5 protein, said nucleic acid comprising IVS1 +198C→ T and/or IVS1 +243G→ C mutation, or

beta-defensin-129 protein, said nucleic acid comprising IVS1 –13_12 in/del CTC mutation.

61. (Currently Amended) The method according to claim 60, wherein the capturing nucleic acid probe is attached or capable of attaching to a solid phase, and comprises a cDNA corresponding to the gene coding a wild-type defensin protein~~, and wherein a detected signal from the solid phase is an indication of the absence of the nucleic acid as defined in claim 49 or 50 in the sample~~.

62. (Original) A transgenic animal which carries a human DNA sequence comprising a nucleotide sequence encoding a variant defensin nucleic acid as defined in claim 49 or 50.

63. (Original) RNA interference methods and models involving a variant nucleotide sequence encoding a variant defensin nucleic acid as defined in claim 49 or 50.

64. (Original) A method for measuring defensin protein expression, production or concentration in human tissues, comprising the steps of:

a) providing a tissue sample taken from a subject to be tested,

b) detecting the expression, production or concentration of defensin protein in said sample,

wherein reduced expression, production or concentration indicates an increased risk of cardiovascular disease in said subject.